



Original Article

A Regional Study on Phytochemical Constituents and Pharmacological Potential of *Hibiscus sabdariffa* Linn. Leaves in Murbad, Maharashtra

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Manuscript ID:

IBMIRJ -2025-021112

Submitted: 07 Oct. 2025

Revised: 20 Oct. 2025

Accepted: 10 Nov. 2025

Published: 30 Nov. 2025

ISSN: 3065-7857

Volume-2

Issue-11

Pp.46-48

November 2025

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Quick Response Code:



Web: <https://ibrj.us>



DOI: [10.5281/zenodo.17659967](https://doi.org/10.5281/zenodo.17659967)

DOI Link:

<https://doi.org/10.5281/zenodo.17659967>



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Abstract

Murbad is one of the tribal talukas of the Thane District in Maharashtra having 206 villages comprising forest area of 35237 hectares and the population of the Murbad tahsil mainly comprises tribal groups that are well-versed in the traditional knowledge of medicinal plants found in the surrounding forest area. The present investigation aims to search into the analysis of the phytochemicals such as alkaloids, Carbohydrates, Saponins, Phytosterols, Phenols, Tannins, Flavonoids, Proteins and its amino acids, Diterpenes, and Glycosides present in the plants of the Hibiscus sabdariffa that is Lal-Ambadi found in the forest areas of the Murbad Tahsil.

Keywords- Lal-Ambadi, alkaloids, Carbohydrates, Saponins, Phytosterols, Phenols, Tannins, Flavonoids, Proteins and its amino acids, triterpenoids, tribal, forest area etc.

Introduction

Red Ambadi is a flowering plant belonging to the *Hibiscus* genus, originally native to Africa, particularly the western regions. In the 1500s and early 1600s, it extended to Asia and the West Indies, where it later established itself naturally in numerous areas. It is either an annual herb or a perennial woody-based subshrub, reaching a height of 7–8 feet. The leaves are deeply three to five lobed 8–15cm in length and alternate to each other on the stems. The flowers measure about 8–10 cm in diameter, ranging in color from white to pale yellow, each petal featuring a dark red spot at its base. The calyx is thick and prominent at the base, initially about 1–2 cm wide and expanding to 3–3.5 cm as the fruit develops. When mature, after approximately six months, it becomes fleshy and turns a deep crimson red.

The *Hibiscus sabdariffa* L. commonly called as Red-Ambadi in Marathi are medical plant belongs to the family of Malvaceae and comprises of different phytochemicals and biochemical compounds which are used for different biological functions upon consumption along with them these are used to treat various human diseases.

In Ayurveda and the traditional Chinese medicine system The use of herbal extracts and dietary supplements for managing various illnesses is well established. The Ambadi plant (*Hibiscus sabdariffa* L., Malvaceae) has been reported to possess diuretic, mild laxative, as well as cardiac and neurological therapeutic properties. The *Hibiscus* has more than 300 species found in tropical and sub-tropical areas in the world and the researcher found that some species of *Hibiscus* has certain medicinal properties of which one is the *Hibiscus sabdariffa* L. which is commonly named as "red sorrel" or "roselle"². In India, Africa and Mexico infusions of the leaves or the calyces of *Hibiscus* have a traditional diuretic, choleric, febrifugal and hypotensive action, reducing the viscosity of the blood and promoting intestinal peristalsis³.

The phytochemicals compounds present in the *Hibiscus Sabdariffa* L. have antioxidant activities like phenol, Phenolic acids and their derivatives, flavonoids, phytic acid, and sterols act as antioxidants, able to neutralize free radicals, bind metal ions, activate antioxidant enzymes, remove alpha-tocopherol radicals, and inhibit oxidase activity.

Phytochemicals are non-nutritive bioactive compounds naturally present in various plant parts, such as flowers, leaves, fruits, roots, bark, spices, and medicinal herbs. In humans, numerous phytochemicals have been found to offer protective and preventive benefits against several degenerative and pathological conditions, including aging, coronary heart disease, Alzheimer's disease, neurodegenerative disorders, atherosclerosis, cataracts, and inflammation.

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How to cite this article:

Rajbhoj, B. G., & Waghmare, J. (2025). A Regional Study on Phytochemical Constituents and Pharmacological Potential of *Hibiscus sabdariffa* Linn. Leaves in Murbad, Maharashtra. *InSight Bulletin: A Multidisciplinary Interlink International Research Journal*, 2(11), 46–48. <https://doi.org/10.5281/zenodo.17659967>

Materials and Methods

Selection of site:- Murbad is one of the tribal talukas of the Thane District in the State of Maharashtra having forest area of 35237 hectares and the population of the said Taluka mainly comprises tribes such as Thakur, Mahadev Koli, Katkari/Kathodi, and ethnic groups such as Kunabi, and Agriculturist. The Medicine men from the Thakur and Katkari tribes are well-versed in medicinal plants. In the past, no ethnoveterinary research was conducted in Murbad tehsil. Therefore, the jurisdiction of Murbad Tahsil is selected for the present investigation.

Plant identification & Collection of Samples:- The plants of the *Hibiscus sabdariffa* L (**Lal-Ambadi**) are identified amongst the forest plants with the help of local tribes and the samples of bark and leaves of *Hibiscus sabdariffa* L (**Lal-Ambadi**) are collected for purpose of the phytochemical studies of the said plant.

Preparation of samples for analysis of Phyto-chemicals:- The leaves of the Red-Ambadi are washed under the running tap water and washed again with distilled water for five-six times and then dried under shade and finally the plant extracts are powdered through mortar and pestle and the powder is filled in clean and dry plastic container.

Preparation of solvent extract for screening of Phyto chemicals: The powdered plant extract were extracted with the 80% aqueous solution of methanol for 6 days. The filtrate is filtered again using muslin cloth to remove fine impurities in the filtrate and the filtrate is filtered again using Whatman No.1 filter paper. Methanol and water were evaporated, and further extracts were then acquired for the analytical purposes.

Screening of Phyto-chemicals in the plants of *Hibiscus sabdariffa* L (**Lal-Ambadi**): Methanolic leaf extracts of the plant are screened for the following phyto-chemicals: alkaloids, flavonoids, saponins, tannins, steroids, proteins and amino acids, triterpenoids, carbohydrate, phenols, phyto-sterols, fixed oils and fats and resins.

Hager's Reagent for alkaloids test: Hager's reagent is added to the extract and it forms a yellow precipitate which shows the presence of alkaloids.

b) A few drops (1–5) of concentrated hydrochloric acid were added to a small amount of the plant extract. The sudden appearance of a red color confirms the presence of flavonoids.

c) **Tannin Test (Gelatin Test):** A few drops of 1% gelatin solution containing sodium chloride were mixed with the plant extract solution. The formation of a white precipitate confirms the presence of tannins.

d) **Test for Saponins (Distilled Water Test):** Five milligrams of the extract were vigorously shaken with 5 ml of distilled water in a test tube and then heated. The appearance of stable foam signifies the presence of saponins.

e) **Test for Steroids (Chloroform Test):** One milligram of the plant extract was added to 10 ml of chloroform along the side of a test tube, An equal volume of concentrated sulfuric acid was then added. The formation of a red color in the upper layer and a yellow layer with green fluorescence beneath indicates the presence of steroids.

Proteins and amino acids are tested by Xanthoproteic test: Few drops of nitric acid (conc. 10%) are added to the extract. Yellow colour: This indicates the presence of proteins.

f) Screening for triterpenoids using conc. sulphuric acid: Chloroform extract, filtered. The filtrate is subsequently mixed with a few drops of concentrated sulfuric acid, shaken thoroughly, and allowed to stand standing if it shows the appearance of a golden yellow colour, then triterpenes are present.

g) Carbohydrate test using Benedict's Reagent - Carbohydrates are present if the Benedict's reagent changes color to blue, orange, or yellow. 2-3ml benedict's reagent is added to 3ml of extract and boiled for 5 minutes.

h) Test for Phenols using ferric chloride solution: A few drops of ferric chloride solution were added to the extract, and the development of a bluish-black color confirmed the presence of phenols.

i) **Test for Phytosterols using Acetic Anhydride Reagent:**

The chloroform extract was filtered, A few drops of acetic anhydride were added to the filtrate, and the mixture was boiled and cooled. Concentrated sulfuric acid was then added, and the formation of a brown ring at the interface confirmed the presence of phytosterols.

Stain Test for Fixed Oils and Fats:

A small portion of the extract was placed and pressed between two filter papers. The appearance of an oily mark confirmed the presence of fixed oils.

k) **Acetone-Water Test:**

The extract was treated with acetone, slightly acidified with a small quantity of water, and shaken. The formation of turbidity indicated the presence of resins.

Results and Discussion

Phytochemical examination of the extracts showed the presence of bioactive compounds known for their medicinal and physiological effects. The analysis confirmed the occurrence of major phytochemicals such as phenols, tannins, saponins, alkaloids, and flavonoids. The relevance of the phytochemicals to drug development is growing because of the large number of bioactive compounds available. Phytochemicals including phytosterols, phenols, saponins, alkaloids, flavonoids and tannins, have been demonstrated to have significant pharmacological potentials in the leaves of *Hibiscus sabdariffa* L. These bioflavonoids, which are often isolated from plants with the help of solvent materials like methanol and water, have many different health benefits that make them useful for the advancement of future pharmaceuticals.

Phytosterols are plant-derived steroid compounds with a structure and function comparable to cholesterol. These plant sterols play an essential role. They are known to exhibit beneficial health effects in humans and animals, particularly in reducing the risk of heart disease, colon cancer, breast cancer, stomach cancer, obesity, and heart attacks. Steroids are typically non-polar or weakly polar molecules and therefore show better solubility in non-polar or less polar solvents such as methanol. Aqueous extracts have been shown to be less powerful dissolving agents of non-polar drugs such as steroids because of their polar nature given that they contain water.

Phenols are among the widely distributed metabolites of plants. The extract had phenolic compounds and these solvents are effective in extraction of the phenol from plant tissues due to their ability to break down plant cell walls and liberate these compounds. Several phenolic compounds may contain structural differences leading to difficulties in extracting them due to the difference in their solubility in these solvents. Phenolic compounds are known to exhibit pharmacological effects such as anti-apoptotic, anti-carcinogenic, anti-inflammatory, anti-aging, anti-atherosclerotic, anti-angiogenic, cell-proliferative, and cardiovascular protective properties. Most phenolics show strong potential for developing medicines targeting disorders linked to oxidative stress and inflammation.

The therapeutic properties of saponins present in the extracts have been reported to include anti-hemorrhagic action (prevention of bleeding), wound and ulcer repair, as well as promoting red blood cell clotting. Additionally, saponins exhibit anti-inflammatory properties, cholesterol-binding capacity, hemolytic activity, bitterness, and antibacterial effects.

Alkaloids are recognized for their significant medicinal effects such as cytotoxic, analgesic, antispasmodic, and antibacterial activities. The extracts contained alkaloid substances, which are basic nitrogen-containing chemical compounds synthesized by various organisms including bacteria, fungi, plants, and animals.

Flavonoids, which are plant metabolites, act as essential signaling compounds and are highly antioxidant in nature. The extracts revealed the presence of flavonoid constituents that form a major class of natural antibiotics. These chemicals play a major role in the defense of plants against different microbes. Flavonoids have been reported to be associated with several pharmacological activities, such as antimicrobial³¹, antioxidant³² and anticancer³³ properties. Flavonoids are known to prevent cell injuries and are therefore, useful in drug compositions directed against oxidative stress and inflammation.

Conclusion

The different types of phytochemicals present in the extracts have a lot of important bioactive roles. These compounds are an excellent source of pharmacological value. Hence, the extracts would be potential leads for the development of the valuable pharmaceuticals. These bioactive phytochemicals present in these Red-Ambadi plants show a great potential for future drug development. Plants that are enriched with various phytochemicals might be a great source of new therapeutic agents for the treatment of a wide variety of diseases. The study highlights the need for more research into the medicinal value of wild plants to fully realize their potential as pharmaceutical products.

Acknowledgments

The author thanks to the tribal, local healers and practitioners, and plant informants for their helpful advice and information, as well as all those who assisted in the fieldwork directly or indirectly, and Sundarrao More College of Arts, Commerce and Science, Poladpur Dist. Raigad-402 303 and Principal Gokhale Education Society, Shrivardhan for continuous encouragement.

Financial support and sponsorship

Nil.

Conflicts of interest

The authors declare that there are no conflicts of interest regarding the publication of this paper.

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